European Journal of Obstetrics & Gynecology and Reproductive Biology 236 (2019) 105–108

Contents lists available at ScienceDirect

European Journal of Obstetrics & Gynecology and Reproductive Biology

journal homepage: www.elsevier.com/locate/ejogrb

Full length article

A clinical study on the difference among proteinuria components in preeclampsia and pregnancies complicated with chronic nephrosis

Xu Zhuang[a,1], Zhuo-cheng Lv[b,1], Jun Shi[a], Jian-hua Lin[a,*]

[a] Department of Obstetrics and Gynecology, Ren Ji Hospital, School of Medicine, Shanghai Jiao Tong University, Shanghai, 200127, China
[b] School of Medicine, Shanghai Jiao Tong University, Shanghai.200025, China

ARTICLE INFO

Article history:
Received 23 December 2018
Received in revised form 3 March 2019
Accepted 5 March 2019

Keywords:
Preeclampsia
Pregnancies complicated with chronic nephrosis
Proteinuria
Proteinuria component analysis

ABSTRACT

Objective: To explore proteinuria components in preeclampsia (PE) and pregnancies complicated with chronic nephrosis (PCCN).

Methods: A case-control study was conducted with 81 PE and 95 PCCN patients and 192 normal pregnancies from April 2016 to March 2018. The results of a 24 h proteinuria test and a proteinuria component analysis (PCA) of all enrolled patients were collected. Statistical analyses of variance and SNK-q were conducted to identify the difference between PE and PCCN in urinary protein components using SPSS 23.0 software. A Pearson test and linear regression were conducted to explore the association between 24 h proteinuria and PCA.

Results: Among the PE, PCCN and control groups, the average values of mAlb(2868.5 ± 3119.3 vs 1586.2 ± 3627.0 vs 21.6 ± 23.6), TRF(252.0 ± 280.5 vs 112.9 ± 164.5 vs 3.1 ± 2.7), α1-MG(40.4 ± 40.7 vs 34.0 ± 38.6 vs 10.3 ± 8.0), β2-MG(1.9 ± 5.1 vs 6.8 ± 15.8 vs 0.9 ± 2.3), and RBP(0.9 ± 1.7 vs 3.1 ± 4.5 vs 0.4 ± 0.7) were significantly different ($P<0.001$). According to the SNK-q test, the average value of mAlb and TRF in the PCCN group is lower than that in the PE group, but higher than the control group ($P < 0.05$). The average value of RBP and β2-MG in the PCCN group was higher than the PE and control groups ($P < 0.05$). The mAlb, TRF, and α1-MG values separately had a significant correlation with the 24 h proteinuria value in PE. The linear regression equation was 24 h proteinuria value = 0.891*mAlb + 5.969*TRF + 1742.378. The mAlb, TRF, α1-MG, β2-MG, and RBP values separately had a significant correlation with the 24 h proteinuria value in PCCN and a linear regression equation of PCCN was as follows: 24 h proteinuria value = 15.148*TRF + 0.571*mAlb.

Conclusions: The proteinuria components of PE and PCCN patients were different in the elevated β2-MG and RBP. The PCA could be a suitable test for qualitative analysis and an antidiastole for PE and PCCN.

Introduction

Preeclampsia (PE) is an idiopathic pregnancy-induced disease which threatens the safety of both the mother and fetus [1]. Proteinuria was once widely considered a necessary criteria of PE's definition [2]. However, since 2013 the diagnosis of PE has been re-defined as a condition accompanied by hypertension with any organ injury, as defined by the updated PE guidelines of the American Congress of Obstetricians and Gynecologists [3], the Society of Obstetricians and Gynecologists of Canada [4], and the Chinese Society of Obstetricians and Gynecologists [5]. Consequently, the clinical diagnostic value of proteinuria was greatly reduced in PE, as there was no clear correlation between proteinuria and pregnancy outcomes [3,4].

Proteinuria as an initial symptom in both PE and chronic nephrosis was common [6]. For nephrologists, components of proteinuria which originate from multiple sources can direct the diagnosis, treatments, and affect the prognosis [7].However, for obstetricians more attention is generally paid to the amount of proteinuria rather than the components.

The proteinuria component analysis (PCA), which includes measuring urinary microalbumin (mAlb), transferrin (TRF) [8], α1-microglobulin (α1-MG), β2-microglobulin (β2-MG) [9], and retinol-binding protein (RBP) [10] has been widely applied for disease localization and diagnosis in Nephrology since 1998. However, there was no report on the application of PCA in pregnancy complicated with chronic nephrosis(PCCN)and PE patients. In the

* Corresponding author at: Department of Obstetrics and Gynecology, Ren Ji Hospital, School of Medicine, Shanghai Jiao Tong University, No. 160, Pujian Road, Shanghai, 200127, China.
E-mail address: linjianhuatg@126.com (J.-h. Lin).
[1] These authors contributed equally to the article.

https://doi.org/10.1016/j.ejogrb.2019.03.007
0301-2115/

current research, patients with PE who showed the presence of proteinuria and pregnancies complicated with chronic nephrosis were examined using the PCA to understand the characteristics of urinary protein for PE and PCCN, facilitating the clinical differential diagnosis of PCA in obstetric applications.

Materials and methods

This study was approved by the Ethics Committee of Ren Ji Hospital, in accordance with the principle of informed consent. An observational study was conducted from April 2016 to March 2018. Eighty-one preeclamptic patients and ninety-five pregnancies accompanied by certain chronic nephrosis history were enrolled. The clinical features were collected.

PE was defined based on the appearance of systolic blood pressure (SBP) ≥140 mmHg and (or) diastolic blood pressure (DBP) ≥ 90 mmHg, accompanied with proteinuria ≥ 0.3 g/24 h after 20 weeks [5]. Also, no renal disease was diagnosed before pregnancy and a renal ultrasound showed a normal image. PE patients were followed until 12 weeks postpartum to exclude the possibility of nephrosis. PE was classified into early onset and late onset by 34 weeks. The mean onset time of the 85 enrolled PE patients was 28.2 ± 5.5 weeks. The proportion of early onset PE was 80.2%(65/81),while that of late onset PE was 19.8%(16/81). Patients were enrolled when PE were clearly diagnosed. The mean detection time of PE patients was 31.1 ± 4.5 weeks.

PCCN was defined as patients with a certain diagnosis of primary chronic glomerular nephrosis before pregnancy with no appearance of PE. Secondary renal diseases, including diabetic nephropathy, lupus nephropathy etc. were excluded from the study. The mean detection time of PCCN patients was 32.6 ± 5.4 weeks.

Moreover, 192 patients with normal pregnancies were simultaneously enrolled as the control group.

The exclusion criteria were defined as the appearance of any of the following: clinical data was incomplete or lost; patients with any other complication; or patients with an unclear diagnosis or diagnosis of PE complicated with chronic nephrosis.

A 24 h proteinuria test was conducted using the pyrogallol red method (total protein UC FS). PCA was conducted by scatter turbidimetry of mAlb (N antiserum to human Alb), TRF (N antiserum to human transferrin), α1-MG (using the N α1-microglobulin kit), β2-MG (using the N latex β2-Microglobulin kit), and immune-turbidimetric assay of RBP (using the retinol binding protein test kit).

Statistical analysis was conducted using SPSS version 23.0 software (IBM, USA). Continuous data were analyzed using the variance (F) test and SNK-q test; a Pearson correlation test and linear regression test were used to detect the association between PCA and 24 h proteinuria value. A P value < 0.050 was considered statistically significant.

Results

Patient information

The study enrolled 176 patients as a study group, including 81 PE patients and 95 PCCN patients. Additionally, 192 patients with normal pregnancies were enrolled as a control group. There was no significant difference between groups in age (F = 0.686, P = 0.504), gravidity (F = 2.336, P = 0.098),and BMI(F = 1.001, P = 0.368), while there was a significant difference between groups in parity (F = 4.185, P = 0.016), delivery weeks(F = 101.538, P < 0.001) and weight of fetus(F = 119.915, P < 0.001) (Table 1).

Characteristics of proteinuria composition

In all 368 patients the same urine sample was examined for the 24 h proteinuria test and the PCA, respectively. These results showed the 24 h proteinuria average value in the PE group was 5802.5 mg ± 4947.6, which was much higher than the PCCN group with 3071.8 mg ± 4092.6 and control group with 143.5 mg ± 64.9 (F = 99.903, P < 0.001). Simultaneously, the average values of mAlb, TRF, α1-MG, β2-MG, and RBP were significantly different among the PE, PCCN and control groups according to variance (F) tests (all P < 0.001) (Table 2).

However, when comparing groups further using the SNK-q test the average value of mAlb and TRF in the PCCN group was lower than the PE group, while higher than the control group (all P < 0.05). However, there was no significant difference in the average value of α1-MG between the PE and PCCN groups (P = 0.094), which was higher than the control group (P < 0.05). Also, the average value of RBP and β2-MG in the PCCN group was higher than the PE and control groups (P < 0.05), while there is no significant difference in the average value of RBP and β2-MG between PE and control groups (P > 0.05) (Graph 1–3).

The correlation and linear regression between PCA and 24 h proteinuria in PE and PCCN

According to the Pearson test, our study showed the mAlb (r = 0.651, P < 0.001), TRF (r = 0.486, P < 0.001), and α1-MG (r = 0.356, P = 0.001) values separately had a significant correlation

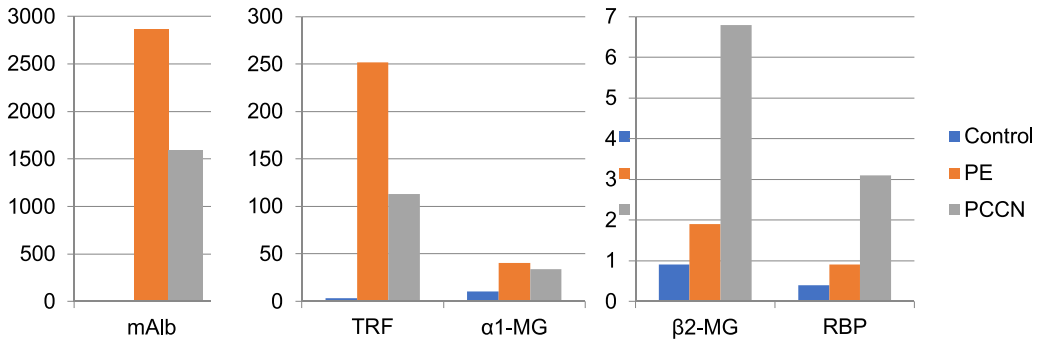
Table 1
Patient information for the groups.

Groups	N	Age (years old)	Gravidity (times)	Parity (times)	BMI (kg/m^2)	Delivery week (weeks)	Weight of fetus (g)	Percentage of FGR (%)
PE	81	30.09 ± 5.26	2.31 ± 1.38	0.42 ± 0.57	25.4 ± 4.1	32.6 ± 4.0	1718 ± 968	44.4(36/81)
PCCN	95	29.58 ± 4.31	1.95 ± 1.25	0.20 ± 0.52	25.8 ± 3.5	35.4 ± 5.2	2591 ± 1015	25.2(24/95)
Control	192	30.32 ± 5.25	2.30 ± 1.45	0.38 ± 0.57	25.0 ± 3.2	38.6 ± 1.0	3217 ± 371	0

Table 2
The difference between groups for the 24 h proteinuria test and PCA results.

Groups	24 h proteinuria value (mg)	mAlb (mg/L)	TRF (mg/L)	α1-MG (mg/L)	β2-MG (mg/L)	RBP (mg/L)
Control	143.5 ± 64.9	21.6 ± 23.6	3.1 ± 2.7	10.3 ± 8.0	0.9 ± 2.3	0.4 ± 0.7
PE	5802.5 ± 4947.6*	2868.5 ± 3119.3*	252.0 ± 280.5*	40.4 ± 40.7*	1.9 ± 5.1	0.9 ± 1.7
PCCN	3071.8 ± 4092.6*,†	1586.2 ± 3627.0*,†	112.9 ± 164.5*,†	34.0 ± 38.6*	6.8 ± 15.8*,†	3.1 ± 4.5*,†
F	99.903	45.124	74.738	42.840	15.405	36.683
P	<0.001	<0.001	<0.001	<0.001	<0.001	<0.001

* Compared to the control group, there was a significant difference.
† Compared to the PE group, there was a significant difference.



Graph 1–3. The difference between groups for the PCA results.

with 24 h proteinuria values, while β2-MG (r = -0.066, P = 0.336) and RBP (r = 0.154, P = 0.171) were not correlated with 24 h proteinuria values. These results were similar to the results in Table 2. Moreover, a linear regression test showed a linear regression equation of 24 h proteinuria can be obtained with a strong positive synthesized correlation of mAlb and TRF in the PE group (r = 0.728, F = 44.023, P < 0.001). The equation for PE was as follows: 24 h proteinuria value = 0.891*mAlb + 5.969*TRF + 1742.378 (Table 3).

On the other hand, according to the Pearson test for the PCCN group, it was shown that the mAlb (r = 0.681, P < 0.001), TRF (r = 0.754, P < 0.001), α1-MG (r = 0.675, P < 0.001), β2-MG (r = 0.301, P = 0.003), and RBP (r = 0.320, P = 0.002) values separately had a significant correlation with 24 h proteinuria values, which were also similar to the results in Table 2. Moreover, a linear regression test showed a linear regression equation of 24 h proteinuria can be obtained with a strong positive synthesized correlation of TRF in the PCCN group (r = 0.896, F = 186.116, P < 0.001). The equation for PCCN was as follows: 24 h proteinuria value = 15.148*TRF + 0.571*mAlb (Table 4).

Discussion

Proteinuria is a common symptom of PE and PCCN, but PE could not be considered until the occurrence of hypertension. However, because renal puncture is not a routine operation due to the high risk during pregnancy, it is often difficult for obstetricians to early identify PE and PCCN through initial symptoms as new-onset proteinuria during pregnancy without an indication of hypertension. In addition, chronic nephritis or other diseases re-diagnosed after a postpartum follow-up of PE were common. Thus, the question remains whether patients with proteinuria greater than or equal to 0.3 g/24 h and high blood pressure have PE, and whether proteinuria is a useless indicator of PE patients' adverse outcomes because the proteinuria components are not well-identified. Therefore, seeking an accurate diagnostic method of early PE was a priority when patients only present with proteinuria.

The difference between proteinuria components in PE and PCCN

Urinary mAlb, TRF, α1-MG, β2-MG, and RBP are the main components of proteinuria, which can be quantified and used to identify the renal injury location by nephrologists. An elevated level of mAlb and TRF indicate glomerular injury, while an elevated level of α1-MG, RBP, and β2-MG indicates a renal tubular absorption injury (Table 5).

In our study, it was shown that the proteinuria composition characteristics of PE and PCCN were significantly different. The levels of urinary mAlb and TRF of the PE group were higher than those of the PCCN and control groups, which may indicate that PE mainly correlates with glomerular injury. This is because PE is a small vascular disease with basic lesions, causing systemic arteriolar spasms. When the lesion occurred in renal arteries (mainly glomerular arteries), it would cause decreased renal blood flow, subsequently leading to leakage of large macromolecule protein such as mAlb and TRF into the urine with a lack of tubular absorption [6,8]. When the glomerular injury is further aggravated,

Table 3
Correlation and linear regression coefficients of 24 h proteinuria values and proteinuria components in the PE group.

24 h proteinuria value of PE		mAlb	TRF	α1-MG	β2-MG	RBP
Correlation coefficient	r	0.651	0.486	0.356	0.011	0.297
	P	<0.001	<0.001	0.001	0.925	0.171
Linear regression coefficient	B	0.891	5.969	0.912	0.984	1.308
	t	6.984	4.207	−0.938	−0.200	0.423
	P	<0.001	<0.001	0.351	0.842	0.647

Table 4
Correlation and linear regression coefficients of 24 h proteinuria values and proteinuria components in the PCCN group.

24 h proteinuria value of PCCN		mAlb	TRF	α1-MG	β2-MG	RBP
Correlation coefficient	r	0.681	0.754	0.675	0.301	0.320
	P	<0.001	<0.001	<0.001	0.003	0.002
Linear regression coefficient	B	0.571	15.148	0.946	1.071	0.087
	t	10.471	12.572	−0.746	1.414	1.727
	P	<0.001	<0.001	0.457	0.161	0.088

Table 5
Correlation between PCA and renal injury location.

Renal injury location	PCA
Glomerular injury	mAlb, TRF
Renal tubular absorption injury	α1-MG, RBP, β2-MG

more proteinuria leaks. Therefore, the proteinuria of PE was related to elevated mAlb and TRF with normal β2-MG and RBP.

However, the pathophysiology of PCCN is different from PE, which influences both glomerular and renal tubule injury. PCCN, as renal parenchymal injury, leads to podocytes injury, subsequently causing the elevated level of mAlb and TRF in the urine. Meanwhile, renal tubules have reabsorption functions which are reflected in the urine composition and urine protein levels. With renal tubular injury, decomposition of main proteins such as RBP and β2-MG is relatively reduced, and these proteins are subsequently exuded through the urine [10]. According to the different pathophysiology, whether the level of urinary β2-MG and RBP are normal or not is a potential indication of antidiastole for PE and PCCN.

The correlation between proteinuria components and 24 h proteinuria

The correlation study found the 24 h proteinuria value in PE was positively correlated with the amount of urinary mAlb, TRF, and α1-MG, while the 24 h proteinuria value in PCCN patients was significantly correlated with all protein components. This may be because PE is a pregnancy-induced disease and its course is often short. Therefore, the renal injury in PE is limited to the glomerulus. Patients with PCCN often have a longer course of the disease, and glomerular injury can often affect renal tubular damage. Therefore, the corresponding protein components of renal tubules in PCCN accounts for a certain proportion in the total amount of 24 h proteinuria. On the other hand, α1-MG, RBP, and β2-MG are small-molecule proteins and are not dominant in proteinuria components, so the correlation coefficient is often low. This is verified in subsequent linear regression equations. From the linear regression equations of PE and PCCN, we found that mALB and TRF are the main independent variables of the equation. These two equations could be used to estimate the 24 h proteinuria values.

Moreover, by comparing the two equations, it was shown the 24 h proteinuria of PE patients was much higher than that of PCCN patients with the same mAlb and TRF value.

Application prospects for PCA

In China, using a urine dipstick for protein measurement is currently regarded as routine in antenatal examinations and is used as an indicator of proteinuria. This screening method has the advantage of being inexpensive and rapid, but it often results in misdiagnosis due to its low accuracy. Such misdiagnoses could result in adverse pregnancy outcomes if the appropriate treatment is delayed. In contrast, when the urine dipstick for protein is positive, a 24 h proteinuria sample is taken as the gold standard for diagnosis. Although the 24 h proteinuria test is very accurate, the method of retaining urine is as inconvenient as it is time consuming.

The PCA may have significant clinical application for proteinuria screening. The time taken for the PCA to be administered is comparable to that of the urine dipstick for measurement of urine protein. Our study found PCA was a good method to characterize and identify PE and PCCN and to quantitatively detect the 24 h proteinuria value in patients with PE or PCCN, as there was a linear regression of the 24 h proteinuria value and the PCA test. Therefore, we believe that the PCA as a routine test is feasible during the entire pregnancy.

Conflicts of interest

None.

Acknowledgements

This study was supported by the National “Twelfth Five-Year” Research and Development Program of China (Grant No. 2014BAI05B05) and Science and Technology Fund Project of Shanghai Jiao Tong University School of Medicine (Grant No. Jyh1704).

References

[1] Iwahashi N, Yamamoto M, Nanjo S, Toujima S, Minami S, Ino K. Down-regulation of indoleamine 2, 3-dioxygenase expression in the villous stromal endothelial cells of placentas with preeclampsia. J Reprod Immunol 2017;119:54–60.
[2] Kintiraki E, Papakatsika S, Kotronis G, Goulis DG, Kotsis V. Pregnancy-Induced hypertension. Hormones (Athens) 2015;14(2):211–23.
[3] American congress of obstetricians and gynecologists. Hypertension in pregnancy. Obstet Gynecol 2013;122:1122–31.
[4] Magee LA, Pels A, Helewa M, Rey E, von Dadelszen P, SOGC Hypertension Guideline Committee. Diagnosis, evaluation, and management of the hypertensive disorders of pregnancy. J Obstet Gynaecol Can 2014;36 (7):575–6.
[5] CSOG Hypertension Guideline Committee. Hypertension in pregnancy guideline(2015). Chin J Obstetr Gynecol 2015;50(10):721–8.
[6] Zhuang X, Chen YY, Zhou Q, Lin JH. Qualitative analysis of diagnostic value of 24-h proteinuria for preeclampsia. Chin Med J (Engl) 2015;128(22):2998–3002.
[7] Ai Z, Xu R, Liu W, Zhou Q, Li B, Huang F, et al. Clinicopathologic features of IgA nephropathy patients with different levels of proteinuria. Clin Nephrol 2016;86(7):35–41.
[8] Dekker GA. Management of preeclampsia. Pregnancy Hypertens 2014;4 (3):246–7.
[9] Mackinnon B, Shakerdi L, Deighan CJ, Fox JG, O'Reilly DS, et al. Urinary transferrin, high molecular weight proteinuria and the progression of renal disease. Clin Nephrol 2003;59(4):252–8.
[10] Guder WG, Ivandic M, Hofmann W. Physiopathology of proteinuria and laboratory diagnostic strategy based on single protein analysis. Clin Chem Lab Med 1998;36(12):935–9.